

**PCT**WORLD INTELLECTUAL PROPERTY ORGANIZATION  
International Bureau

## INTERNATIONAL APPLICATION PUBLISHED UNDER THE PATENT COOPERATION TREATY (PCT)

<b>(51) International Patent Classification <sup>4</sup> :</b> A61K 39/02, 39/08, 39/10 A61K 39/11, 35/24	<b>A1</b>	<b>(11) International Publication Number:</b> WO 90/01335 <b>(43) International Publication Date:</b> 22 February 1990 (22.02.90)
<b>(21) International Application Number:</b> PCT/AU89/00328 <b>(22) International Filing Date:</b> 2 August 1989 (02.08.89)  <b>(30) Priority data:</b> PI 9613 2 August 1988 (02.08.88) AU PJ 3837 21 April 1989 (21.04.89) AU  <b>(71)(72) Applicant and Inventor:</b> BORODY, Thomas, Julius [AU/AU]; 144 Great North Road, Five Dock, NSW 2046 (AU).  <b>(74) Agent:</b> SPRUSON & FERGUSON; G.P.O. Box 3898, Syd- ney, NSW 2001 (AU).  <b>(81) Designated States:</b> AT (European patent), AU, BE (Euro- pean patent), CH (European patent), DE (European pa- tent), FR (European patent), GB (European patent), IT (European patent), LU (European patent), NL (Euro- pean patent), SE (European patent), US.		<b>Published</b> <i>With international search report.</i>
<b>(54) Title:</b> TREATMENT OF GASTRO-INTESTINAL DISORDERS  <b>(57) Abstract</b>  A method of treating chronic disorders associated with the presence of abnormal microflora or an abnormal distribution of microflora in the gastrointestinal tract. The method involves the removal of at least a portion of the host's existing enteric microflora and the substitution of an effective amount of predetermined microflora. Pharmaceutical compositions which comprise viable microorganisms in a composition resembling the host's normal healthy faecal flora are also disclosed.		

*FOR THE PURPOSES OF INFORMATION ONLY*

Codes used to identify States party to the PCT on the front pages of pamphlets publishing international applications under the PCT.

AT	Austria	ES	Spain	MG	Madagascar
AU	Australia	FI	Finland	ML	Mali
BB	Barbados	FR	France	MR	Mauritania
BE	Belgium	GA	Gabon	MW	Malawi
BF	Burkina Faso	GB	United Kingdom	NL	Netherlands
BG	Bulgaria	HU	Hungary	NO	Norway
BJ	Benin	IT	Italy	RO	Romania
BR	Brazil	JP	Japan	SD	Sudan
CA	Canada	KP	Democratic People's Republic of Korea	SE	Sweden
CF	Central African Republic	KR	Republic of Korea	SN	Senegal
CG	Congo	LI	Liechtenstein	SU	Soviet Union
CH	Switzerland	LK	Sri Lanka	TD	Chad
CM	Cameroon	LU	Luxembourg	TG	Togo
DE	Germany, Federal Republic of	MC	Monaco	US	United States of America
DK	Denmark				

- 1 -

TREATMENT OF GASTRO-INTESTINAL DISORDERSTechnical Field

The present invention relates to methods of treating diseases in mammals, in particular to the treatment of chronic disorders associated with the presence of abnormal or an abnormal distribution of microflora in the gastrointestinal tract. The invention also relates to pharmaceutical compositions suitable for the treatment of such disorders.

Background Art

There are large numbers of patients suffering from gastro-intestinal symptoms referable to the lower small bowel and large bowel which to date have eluded explanation. These disorders include irritable bowel syndrome (IBS) or spastic colon, idiopathic ulcerative colitis, mucous colitis, collagenous colitis, Crohn's disease, inflammatory bowel disease in general, microscopic colitis, antibiotic-associated colitis, idiopathic or simple constipation, diverticular disease, and AIDS enteropathy. Pathophysiology of these disorders eludes logical explanation in spite of decades of research and millions of dollars of research funds. A common underlying factor shared by all these disorders observed by the present inventor is their onset following some extraneous invading infection. In all the disorders, the infection cannot be demonstrated due to our inability to detect infecting agents whose cultural characteristics are unknown to medical science.

Circumstantial evidence which suggests that these disorders are "infection-related" includes:

- (a) onset following a gastro-intestinal infection which failed to completely resolve;
- (b) transient improvement with use of certain antibiotics, but recurrence upon cessation of antibiotics;
- (c) transient improvement following orthostatic lavage prior to colonoscopy and;
- (d) transient symptom improvement with use of "colonic" irrigation.

It is impractical to use long-term antibiotic therapy (with its associated complications) in such patients since cure is not obtained with its use. Furthermore, chronic gut infections with recognised, specific pathogens such as Clostridium difficile, Yersinia enterocolitica or Campylobacter jejuni/coli are not eradicated with antibiotics. Some previous attempts have been made to alter the enteric microflora in order to eradicate such chronic infections. These measures nevertheless

**SUBSTITUTE SHEET**

- 2 -

indicate that alteration of bacterial flora may effect dramatic clinical improvement in conditions characterized by chronic, resistant enterocolitic infection. However there remain many chronic disorders of uncertain aetiology or causation, which are resistant to cure by current therapeutic techniques.

#### Objects of the Invention

It is thus an object of the present invention to provide novel methods of treating various disease states related to the presence of "abnormal" microflora in the gastrointestinal tract.

#### Disclosure of the Invention

The present invention recognises chronic infection/infestation as the underlying pathological process in a wide range of chronic disorders such as irritable bowel syndrome, particularly when characterised by chronic abdominal pain, bloating, or excessive flatulence, together with chronic diarrhoea or alternating constipation/diarrhoea, and also in spastic colon, mucous colitis, collagenous colitis, ulcerative colitis, Crohn's colitis, microscopic colitis, idiopathic inflammatory bowel disease, antibiotic-associated colitis, idiopathic or simple constipation, diverticular disease and AIDS enteropathy.

The invention has also been found to relate to other gastrointestinal disorders of unexplained aetiology such as polyposis coli and colonic polyps, which may well be influenced by the local bowel microflora.

In addition the present invention also provides a method of treatment of chronic gastrointestinal infections with specific microorganisms such as Clostridium difficile, Yersinia spp, Campylobacter spp, Aeromonas spp, E. coli, Cryptosporidium spp, Amoebae, Giardia and even chronic viral infections, and of small bowel bacterial overgrowth.

The present invention furthermore, recognises the close association between the intestine and liver disease, and the intestine and migraines and chronic fatigue syndrome, and possibly other neurological syndromes such as, multiple sclerosis, amyotrophic lateral sclerosis, myasthenia gravis, Parkinson's disease, Alzheimers disease and other degenerative disorders. Hence, it is proposed that a considerable proportion of currently unexplained diseases of the liver and nervous system of unknown aetiology may be explicable by the chronic storage of pathogens within the small/large intestine and the subsequent passage of antigenic material and pathogenic TOXINS into the portal system (liver damage) or systemic circulation (neurological conditions). Specifically, such hepato/biliary

- 3 -

system disorders as primary biliary cirrhosis, primary sclerosing cholangitis, fatty liver of unknown aetiology, or cryptogenic cirrhosis, may be secondary to chronic pathogen carrier state in the intestine.

The links between the intestine and joint disease are also recognised. Joint diseases such as rheumatoid arthritis, the non-rheumatoid arthritides including, ankylosing spondylitis, and Reiter's syndrome, may also be causally related to a chronic intestinal carrier state, as may other syndromes with an immune mediated component such as glomerulonephritis, haemolytic uraemic syndrome, juvenile diabetes mellitus, Behcet's syndrome, coeliac disease and dermatitis herpetiformis. Similarly, syndromes with an immune complex mediated component; such as scleroderma, systemic lupus erythematosus, mixed cryoglobulinaemia, polyarteritis, familial Mediterranean fever, amyloidosis, and the various presentations of such syndromes, together with such "idiopathic" states as chronic urticaria, may be manifestations of variations of immune regulated responses to related bowel-origin pathogens chronically shedding their antigen(s) into the circulation. Other chronic conditions such as acne, and chronic idiopathic pseudo-obstructive syndrome, may well be influenced by similar mechanisms.

For many of these syndromes present therapy offers only palliation of symptoms and/or the induction of remission of the disease process but not cure. The present inventor therefore recognised the need to find a curative therapy for these wide ranging disease processes associated with considerable morbidity.

Thus according to a first embodiment of the invention there is provided a method of treatment or prophylaxis of a chronic disorder associated with the presence in the gastrointestinal tract of a host of abnormal or an abnormal distribution of microflora for said host, which method comprises the removal of at least a portion of the host's existing enteric microflora and the substitution of an effective amount of predetermined microflora.

In its preferred form the treatment should effect a cure of the symptoms of such disorders. The change of flora is preferably as "near-complete" as possible and the flora is replaced by viable organisms which will crowd out any remaining, original flora.

The method of the present invention is applicable to animals in general in particular humans and economically significant domestic animals.

In the case of humans, the present invention encompasses methods of

**SUBSTITUTE SHEET**

- 4 -

treatment of chronic disorders associated with the presence of abnormal enteric microflora. Such disorders include but are not limited to those conditions in the following categories:

i) gastro-intestinal disorders including irritable bowel syndrome or spastic colon, ulcerative colitis, mucous colitis, collagenous colitis, Crohn's disease, inflammatory bowel disease, microscopic colitis, antibiotic associated colitis, idiopathic or simple constipation, diverticular disease, AIDS enteropathy, small bowel bacterial overgrowth, coeliac disease, polyposis coli, colonic polyps, chronic idiopathic pseudo obstructive syndrome;

ii) chronic gut infections with specific pathogens including bacteria, viruses, fungi and protozoa;

iii) liver disorders such as primary biliary cirrhosis, primary sclerosing cholangitis, fatty liver or cryptogenic cirrhosis;

iv) joint disorders such as rheumatoid arthritis, non-rheumatoid arthritides, ankylosing spondylitis, and Reiter's syndrome;

v) immune mediated disorders such as glomerulonephritis, haemolytic uraemic syndrome, juvenile diabetes mellitus, mixed cryoglobulinaemia, polyarteritis, familial Mediterranean fever, amyloidosis, scleroderma, systemic lupus erythematosus, and Behcet's syndrome;

vi) neurological syndromes such as migraine, multiple sclerosis, amyotrophic lateral sclerosis, myasthenia gravis, chronic fatigue syndrome, Parkinson's disease, Alzheimers disease and other degenerative disorders;

vii) dermatological conditions such as, chronic urticaria, acne, dermatitis herpetiformis and vasculitic disorders;

The above disorders are all characterised by their response to treatment with the method of the present invention.

Typically the change in enteric flora comprises:

- (a) substantially complete removal of existing enteric flora, and
- (b) introduction of an array of predetermined flora into the gastro-intestinal system, and thus in a preferred form the method of treatment comprises substantially completely changing enteric flora in patients requiring such treatment.

Furthermore, in some of these disorders a short course of antibiotics may be required to rid tissue-invasive pathogens originating in the bowel lumen. For example, in Crohn's disease, anti-tuberculosis therapy may be required for six to twelve weeks before the bowel is cleared out and the flora content exchanged for a predetermined flora.

**SUBSTITUTE SHEET**

- 5 -

Preferably the removal of existing enteric flora is effected by lavage of the gastro-intestinal tract. This can be effected by methods known to those skilled in the art such as ingestion of lavage solutions such as orthostatic salt and polyethylene glycol solution, enemas or small bowel intubation and lavage.

Turning to the introduction of an array of predetermined flora into the gastro-intestinal system, this can be effected by enemas or per-colonoscopy, via intubation of the small bowel using for example a large bore catheter equipped with distal balloon to effect rapid passage down the jejunum, or via the oral route with enteric-coated capsules.

The predetermined array of normal bowel microflora preferably comprises a composition of fresh homologous faeces, equivalent freeze-dried and reconstituted faeces or a "synthetic" faecal composition.

In a preferred form the synthetic faecal composition comprises a preparation of viable flora which preferably in proportional content, resembles normal healthy human faecal flora. Suitable microorganisms may be selected from the following; Bacteroides, Bifidobacterium, Eubacteria, Fusobacteria, Propionibacteria, Lactobacilli, anaerobic cocci, Ruminococcus, E. coli, Gemmiger, Clostridium, Desulfomonas, species and, more specifically, bacteria selected from Table 1. Preferably fungi are also present such as Monilia.

According to a second embodiment of the invention there is provided a pharmaceutical composition useful for the treatment and/or prophylaxis of chronic disorders associated with the presence in the gastrointestinal tract of a host of abnormal or an abnormal distribution of microflora for said host, which composition comprises a selection of viable microorganisms in appropriate proportions such that the composition substantially resembles the host's normal healthy, faecal flora.

In practice suitable microorganisms include those selected from Bacteroides, Bifidobacterium, Eubacteria, Fusobacteria, Propionibacteria, Lactobacilli, anaerobic cocci, Ruminococcus, E. coli, Gemmiger, Clostridium, Desulfomonas, and Monilia species, and more specifically from those set out in Table 1.

In a preferred form the composition comprises a liquid culture of Bacteroides and E. coli.

The pharmaceutical composition of the present invention is preferably lyophilised, pulverised and powdered. It may then be infused, dissolved such as in saline, as an enema. Alternatively the powder may be encapsulated as enteric-coated capsules for oral administration. As a

**SUBSTITUTE SHEET**

- 6 -

powder it can preferably be provided in a palatable form for reconstitution for drinking. The composition can be combined with other adjuvants such as antacids to dampen bacterial inactivation in the stomach. Acid secretion in the stomach could also be pharmacologically suppressed using H<sub>2</sub>-antagonists or omeprazole. The powder may be reconstituted also to be infused via naso-duodenal infusion.

The present composition is therefore preferably in the form of:

- i) an enema composition which can be reconstituted with an appropriate diluent;
- ii) enteric-coated capsules, or
- iii) powder for reconstitution with an appropriate diluent for naso-enteric infusion or colonoscopic infusion, or
- iv) powder for reconstitution with appropriate diluent, flavouring and gastric acid suppression agent for oral ingestion.

Furthermore the present invention also relates to the treatment of animals, in particular to the treatment of gastrointestinal disorders in economically important domestic animals, such as cattle, sheep, horses, pigs, goats etc. The method of the present invention has been found to be especially useful in the treatment of the various forms of necrotising enterocolitis which can be a major problem in animal stocks.

Obviously in the treatment of animals the appropriate composition of microflora will vary according to the species being treated and the constituent normal flora known to inhabit the gut. Thus the composition according to the invention would comprise, a preparation of viable flora which preferably in proportional content, resembles the normal healthy faecal flora of the species involved. The compositions may be prepared in any of the forms already described and administered accordingly.

#### Best Method of Performing the Invention

Typically the method of the invention is applicable to a patient suffering from a chronic disorder associated with the presence of abnormal microflora in the gastrointestinal tract such as irritable bowel syndrome.

In the practice of the invention the patients existing enteric flora is removed by gastrointestinal lavage effected by ingestion of about 3 litres of a balanced salt solution with polyethylene glycol. Lavage is continued until the removal of the existing flora is as near complete as possible.

A composition of predetermined flora in the form of a liquid culture of Bacteroides and E. coli is then infused into the patient per colonoscope

**SUBSTITUTE SHEET**



- 7 -

in an amount sufficient to replace the removed flora, and reverse the disease process. Alternatively fresh homologous faeces obtained from a disease screened donor are liquefied and mixed with unprocessed bran. The mixture is then homogenised anaerobically under CO<sub>2</sub> cover and infused into the patient per colonoscope.

Cure or remission of symptoms is then monitored subjectively and by assessment of stool frequency or other appropriate criteria.

Using liquid cultures of Bacteroides and E. coli the inventor has achieved total reversal of colitis, irritable bowel syndrome and constipation.

As indicated in the method of treatment aspect of the invention, a preparatory course of appropriate antibiotics may be used. For example, Septrin for chronic yersiniasis, Metronidazole for ulcerative colitis, anti-TB therapy in Crohn's disease, or Vancomycin in chronic Clostridium difficile infestations.

Table 1

% of florab	Organism(s)
11.8(0.90)	<i>Bacteroides fragilis</i> ss. <i>vulgatus</i>
9.9(0.83)	<i>Eubacterium aerofaciens</i>
8.9(0.78)	<i>Bacteroides fragilis</i> ss. <i>thetaiotaomicron</i>
6.6(0.68)	<i>Peptostreptococcus productus</i> II
6.0(0.64)	<i>Bacteroides fragilis</i> ss. <i>distasonis</i>
4.4(0.55)	<i>Fusobacterium prausnitzii</i>
3.5(0.49)	<i>Coprococcus eutactus</i>
3.0(0.45)	<i>Eubacterium aerofaciens</i> III
2.8(0.44)	<i>Peptostreptococcus productus</i> I
2.7(0.43)	<i>Ruminococcus bromii</i>
2.6(0.43)	<i>Bifidobacterium adolescentis</i>
2.2(0.39)	<i>Gemmiger formicilis</i> , <i>Bifidobacterium longum</i>
2.1(0.38)	<i>Eubacterium siraeum</i>
1.8(0.35)	<i>Ruminococcus torques</i>
1.7(0.34)	<i>Eubacterium rectale</i> III-H
1.6(0.33)	<i>Eubacterium rectale</i> IV; <i>Eubacterium eligens</i>
1.5(0.32)	<i>Bacteroides eggerthii</i>
1.4(0.31)	<i>Clostridium leptum</i>
1.3(0.29)	<i>Bacteroides fragilis</i> ss. <i>a</i>
1.2(0.29)	<i>Eubacterium bifforme</i>

SUBSTITUTE SHEET

- 8 -

Table 1 (cont)

0.91(0.25)	<i>Bifidobacterium infantis</i>
0.84(0.24)	<i>Eubacterium rectale</i> III-F
0.57(0.20)	<i>Coprococcus comes</i> , <i>Bacteroides capillosus</i>
0.50(0.18)	<i>Ruminococcus albus</i> , <i>Eubacterium formicigenerans</i> , <i>Eubacterium hallii</i> , <i>Eubacterium ventriosum</i> I, <i>Fusobacterium russii</i>
0.43(0.17)	<i>Ruminococcus obeum</i> , <i>Eubacterium rectale</i> II, <i>Clostridium ramosum</i> I, <i>Lactobacillus leichmanii</i>
0.36(0.16)	<i>Ruminococcus callidus</i> , <i>Butyrivibrio crossotus</i>
0.30(0.14)	<i>Acidaminococcus fermentans</i> , <i>Eubacterium ventriosum</i> , <i>Bacteroides fragilis</i> ss. <i>fragilis</i> , <i>Bacteroides</i> AR
0.23(0.12)	<i>Coprococcus catus</i> , <i>Eubacterium hadrum</i> , <i>E. cylindroides</i> , <i>E. ruminantium</i> , <i>Eubacterium</i> CH-1, <i>Staphylococcus epidermidis</i>
0.17(0.10)	<i>Peptostreptococcus</i> BL, <i>Eubacterium limosum</i> , <i>Bacteroides praeacutus</i> , <i>Bacteroides</i> L, <i>Fusobacterium mortiferum</i> I, <i>F. naviforme</i> , <i>Clostridium innocuum</i> , <i>C. ramosum</i> , <i>Propionibacterium acnes</i> , <i>Ruminococcus flavefaciens</i>
0.10(0.08)	<i>Ruminococcus</i> AT, <i>Peptococcus</i> AU-1, <i>Eubacterium</i> AG, -AK, -AL, -AL-1, -AN; <i>Bacteroides fragilis</i> ss. <i>ovatus</i> , -ss. d, -ss. f; <i>Bacteroides</i> L-1, L-5; <i>Fusobacterium nucleatum</i> , <i>F. mortiferum</i> , <i>Escherichia coli</i> , <i>Streptococcus morbillorum</i>
0.05(0.05)	<i>Peptococcus magnus</i> , <i>Peptococcus</i> G, -AU-2; <i>Streptococcus intermedius</i> , <i>Ruminococcus lactaris</i> , <i>Ruminococcus</i> CO, <i>Gemmiger</i> X, <i>Coprococcus</i> BH, -CC; <i>Eubacterium tenue</i> , <i>Eubacterium ramulus</i> , <i>Eubacterium</i> AE, -AG-H, -AG-M, -AJ, -BW-1; <i>Bacteroides clostridiiformis</i> ss. <i>clostridiiformis</i> , <i>B. coagulans</i> , <i>B. oralis</i> , <i>B. ruminicola</i> ss. <i>brevis</i> , -ss. <i>ruminicola</i> , <i>Bacteroides splanchnicus</i> , <i>Desulfomonas pigra</i> , <i>Bacteroides</i> L-4, -W-1; <i>Fusobacterium</i> H, <i>Lactobacillus</i> G, <i>Succinivibrio</i> A

<sup>b</sup> The percentage of the fecal population (the standard deviation of the estimate is given in parentheses).

The invention will now be further described with reference to the following non-limiting examples.

**SUBSTITUTE SHEET**

- 9 -

Example 1

A 28 year old male presented with bloody diarrhoea, 7 to 12 times per day, weight loss, anaemia, arthritis, marked liver-function blood test abnormalities, colonoscopic findings of "pancolitis" and histologic diagnosis of active chronic colitis. Following two years of "standard" anti-colitis therapy he improved markedly. The anaemia was corrected, he gained weight and his liver functions improved somewhat. However, he continued to experience three to four stools per day in spite of 9 x 500 mg salazopyrin tablets per day and second-nightly steroid enemas (Predsol Enemas). Furthermore, liver function tests remained elevated. Following one week of metronidazole tablets 200 mg x 4 per day, and oral ingestion of three litres balanced salt solution with polyethylene glycol (GLYCOPREP), he received fresh, liquefied homogenized human donor faeces from a disease-screened donor were infused into the patient. Prior to infusion unprocessed bran was added to the faeces and homogenization was carried out anaerobically under CO<sub>2</sub> cover.

Following infusion, the patient has remained well and off all treatment for three months to date. Liver function tests have returned to normal. He has one formed stool per day and has begun to put on weight. He has no arthralgia.

Example 2

Similar methods were used to treat 55 patients suffering with either constipation, diarrhoea, abdominal pain, ulcerative colitis or Crohn's disease. Patients were treated when other forms of therapy had failed to control their symptoms. Following bowel flora alteration 20 patients were deemed "cured", 9 improved, while 26 failed to improve. The following cases illustrate "cures" using the method of the invention over a follow up period of 1 to 12 months.

Case 1 Chronic constipation

A 31 year old female who had a history of chronic constipation commencing at birth with a frequency of once per week without laxatives. Following bowel flora alteration she has had daily or second stool frequency, without resort to laxatives and remains well at six month follow-up.

Case 2 Chronic Pain-Predominant IBS

This 21 year old female was thoroughly investigated over a four year period for severe colicky abdominal pain, requiring frequent admissions and narcotic pain relief. Multiple investigations included endoscopy,

**SUBSTITUTE SHEET**

- 10 -

laparoscopy and laparotomy. Five days following alteration of her bowel flora she was pain free and remains pain free at four months follow-up.

Case 3      Chronic Diarrhoea-Predominant IBS

A 35 year old female presented with a history of several years of diarrhoea (2-10/d) and associated abdominal pain. Her only abnormal investigation was +ve stool latex test for clostridium difficile toxin. This failed to clear after a course of Vancomycin and she underwent bowel flora alteration with resolution of her chronic diarrhoea. Diarrhoea and pain have not recurred at one month follow-up.

Case 4      Chronic Ulcerative Colitis

This 45 year old man presented with an 18 month history of ulcerative colitis and elevated liver transaminases. Pancolitis was confirmed on colonoscopy. Sulfasalazine caused a rash while olsalazine gave inadequate relief. The patient underwent exchange of bowel flora improving adequately enough to come off treatment within days. At three months he continues to feel well, has no diarrhoea and is on no medication. His liver transaminases have returned to normal. Colonoscopy is now normal and mucosal biopsies are now normal.

Case 5      Crohn's Disease

This 31 year old man was admitted to hospital with small bowel obstruction subsequently shown on small bowel enema to be Crohn's disease of the terminal ileum. This was confirmed on terminal ileal biopsy. In spite of prednisone and sulfasalazine, hypoproteinaemia with ankle oedema was prominent presumably due to protein losing enteropathy. Three days following bowel flora alteration his ankle oedema and serum proteins returned to normal. He remains free of symptoms, off therapy 4 months later.

Case 6      Irritable bowel/chronic fatigue syndrome

This 42 year old woman presented with symptoms of irritable bowel syndrome characterised by diarrhoea and pain, and also complained of profound tiredness. Faecal infusion reversed the bowel symptoms. In addition the profound tiredness suddenly disappeared. Similar results were obtained in 4 related cases.

SUBSTITUTE SHEET

- 11 -

TREATMENT OF GASTRO-INTESTINAL DISORDERS

## CLAIMS

1. A method for the treatment and/or prophylaxis of a chronic disorder associated with the presence in the gastrointestinal tract of a host of abnormal or an abnormal distribution of microflora for said host, which method comprises the removal of at least a portion of the host's existing enteric microflora and the substitution of an effective amount of predetermined microflora.

2. The method of claim 1 wherein the existing enteric microflora is substantially completely removed.

3. The method of claim 1 or 2 wherein the predetermined microflora is in the form of a composition of fresh homologous faeces, equivalent freeze-dried and reconstituted faeces or a synthetic faecal composition.

4. The method of claim 3 wherein the synthetic faecal composition comprises a preparation of a selection of viable microorganisms in appropriate proportions such that the preparation substantially resembles the host's normal, healthy, faecal flora.

5. The method of claim 3 or claim 4 wherein the predetermined flora comprises at least two microorganisms selected from the group consisting of:

Bacteroides, Bifidobacterium, Eubacteria, Fusobacteria, Propionibacteria, Lactobacilli, anaerobic cocci, Ruminococcus, E. coli, Gemmiger, Clostridium, Desulfomonas, and Monilia species.

6. The method of claim 5 wherein the predetermined microflora comprises at least two of the microorganisms listed in the following group:

Bacteroides fragilis ss. vulgatus, Eubacterium aerofaciens, Bacteroides fragilis ss. thetaiotaomicron, Peptostreptococcus productus II, Bacteroides fragilis ss. distasonis, Fusobacterium prausnitzii, Coprococcus eutactus, Eubacterium aerofaciens III, Peptostreptococcus productus I, Ruminococcus bromii, Bifidobacterium adolescentis, Gemmiger formicilis, Bifidobacterium longum, Eubacterium siraeum, Ruminococcus torques, Eubacterium rectale III-H, Eubacterium rectale IV, Eubacterium eligens, Bacteroides eggerthii, Clostridium leptum, Bacteroides fragilis ss. a, Eubacterium biforme, Bifidobacterium infantis, Eubacterium rectale III-F, Coprococcus comes, Bacteroides capillosus, Ruminococcus albus, Eubacterium formicigenerans, Eubacterium hallii, Eubacterium ventriosum I, Fusobacterium russii, Ruminococcus obeum, Eubacterium rectale II, Clostridium ramosum I, Lactobacillus leichmanii, Ruminococcus callidus, Butyrivibrio crossotus, Acidaminococcus fermentans, Eubacterium ventriosum,

SUBSTITUTE SHEET

- 12 -

Bacteroides AR, Bacteroides fragilis, ss. fragilis, Coprococcus catus, Eubacterium hadrum, E. cylindroides, E. ruminantium, Eubacterium CH-1, Staphylococcus epidermidis, Peptostreptococcus BL, Eubacterium limosum, Bacteroides praeacutus, Bacteroides L, Fusobacterium mortiferum I, F. naviforme, Clostridium innocuum, C. ramosum, Propionibacterium acnes, Ruminococcus flavefaciens, Ruminococcus AT, Peptococcus AU-1, Eubacterium AG, -AK, -AL, -AL-1, -AN; Bacteroides fragilis ss. ovatus, -ss. d, -ss. f; Bacteroides L-1, L-5; Fusobacterium nucleatum, F. mortiferum, Escherichia coli, Streptococcus morbillorum, Peptococcus magnus, Peptococcus G, -AU-2; Streptococcus intermedius, Ruminococcus lactaris, Ruminococcus CO, Gemmiger X, Coprococcus BH, -CC; Eubacterium tenue, Eubacterium ramulus, Eubacterium AE, -AG-H, -AG-M, -AJ, -BW-1, Bacteroides clostridiiformis ss. clostridiiformis, B. coagulans, B. oralis, B. ruminicola ss. brevis, -ss. ruminicola, Bacteroides splanchnicus, Desulfomonas pigra, Bacteroides L-4, -W-1; Fusobacterium H, Lactobacillus G, and Succinivibrio A.

7. The method of any one of claims 3, 5 or 6 wherein the predetermined microflora is in the form of a liquid culture of Bacteroides and E. coli.

8. The method of any one of claims 1 to 7 wherein the host is a human.

9. The method of any one of claims 1 to 8 wherein the chronic disorder is a gastrointestinal disorder such as;

irritable bowel syndrome, spastic colon, ulcerative colitis, mucous colitis, collagenous colitis, Crohn's disease, inflammatory bowel disease, microscopic colitis, antibiotic associated colitis, idiopathic or simple constipation, diverticular disease, AIDS enteropathy, small bowel bacterial overgrowth, coeliac disease, polyposis coli, colonic polyps, chronic idiopathic pseudo obstructive syndrome and chronic enteric infections.

10. The method of claim 9 wherein the chronic enteric infection is caused by a bacteria, virus, protozoa or fungi.

11. The method of any one of claims 1 to 8 wherein the chronic disorder is a liver disorder such as primary biliary cirrhosis, primary sclerosing cholangitis, fatty liver or cryptogenic cirrhosis.

12. The method of any one of claims 1 to 8 wherein the chronic disorder is a joint disorder such as rheumatoid arthritis, a non-rheumatoid arthritides, ankylosing spondylitis, or Reiter's syndrome.

**SUBSTITUTE SHEET**

- 13 -

13. The method of any one of claims 1 to 8 wherein the chronic disorder is an immune mediated disorder such as glomerulonephritis, haemolytic uraemic syndrome, juvenile diabetes mellitus, mixed cryoglobulinaemia, polyarteritis, familial Mediterranean fever, amyloidosis, scleroderma, systemic lupus erythematosus, or Behcet's syndrome.

14. The method of any one of claims 1 to 8 wherein the chronic disorder is a neurological disorder such as migraine, multiple sclerosis, amyotrophic lateral sclerosis, myasthenia gravis, chronic fatigue syndrome, Parkinson's disease, or Alzheimers disease.

15. The method of any one of claims 1 to 8 wherein the chronic disorder is a dermatological disorder such as chronic urticaria, acne, dermatitis herpetiformis or a vasculitic disorder.

16. The method of any one of claims 1 to 7 wherein the host is selected from the group consisting of; cows, sheep, horses, goats and pigs.

17. The method of claim 16, wherein the chronic disorder is necrotising enterocolitis.

18. The method of any one of claims 1 to 17 wherein the removal of existing flora is carried out by gastrointestinal lavage with orthostatic salt solution and polyethylene glycol solution.

19. The method of claim 18 wherein gastrointestinal lavage is effected by enemata, oral ingestion or per enteral infusion.

20. The method of any one of claims 1 to 19 wherein the predetermined flora is introduced into the patients gastrointestinal system by ingestion or by per enteral infusion.

21. The method of any one of claims 1 to 20, further comprising the administration of an effective amount of at least one antibiotic prior to the removal of the existing flora.

22. A pharmaceutical composition useful for the treatment and/or prophylaxis of chronic disorders associated with the presence in the gastrointestinal tract of a host of abnormal or an abnormal distribution of microflora for said host, which composition comprises a selection of viable microorganisms in appropriate proportions such that the composition substantially resembles the host's normal healthy, faecal flora.

23. The composition of claim 22 wherein the microorganisms are selected from the group consisting of:

Bacteroides, Bifidobacterium, Eubacteria, Fusobacteria, Propionibacteria, Lactobacilli, anaerobic cocci, Ruminococcus, E. coli, Gemmiger, Clostridium, Desulfomonas, and Monilia species.

SUBSTITUTE SHEET

- 14 -

24. The composition of claim 23 wherein the microorganisms are selected from those listed in the following group:

*Bacteroides fragilis* ss. *vulgatus*, *Eubacterium aerofaciens*, *Bacteroides fragilis* ss. *thetaiotaomicron*, *Peptostreptococcus productus* II, *Bacteroides fragilis* ss. *distasonis*, *Fusobacterium prausnitzii*, *Coprococcus eutactus*, *Eubacterium aerofaciens* III, *Peptostreptococcus productus* I, *Ruminococcus bromii*, *Bifidobacterium adolescentis*, *Gemmiger formicilis*, *Bifidobacterium longum*, *Eubacterium siraeum*, *Ruminococcus torques*, *Eubacterium rectale* III-H, *Eubacterium rectale* IV, *Eubacterium eligens*, *Bacteroides eggerthii*, *Clostridium leptum*, *Bacteroides fragilis* ss. *a*, *Eubacterium biforme*, *Bifidobacterium infantis*, *Eubacterium rectale* III-F, *Coprococcus comes*, *Bacteroides capillosus*, *Ruminococcus albus*, *Eubacterium formicigenerans*, *Eubacterium hallii*, *Eubacterium ventriosum* I, *Fusobacterium russii*, *Ruminococcus obeum*, *Eubacterium rectale* II, *Clostridium ramosum* I, *Lactobacillus leichmanii*, *Ruminococcus callidus*, *Butyrivibrio crossotus*, *Acidaminococcus fermentans*, *Eubacterium ventriosum*, *Bacteroides* AR, *Bacteroides fragilis*, ss. *fragilis*, *Coprococcus catus*, *Eubacterium hadrum*, *E. cylindroides*, *E. ruminantium*, *Eubacterium* CH-1, *Staphylococcus epidermidis*, *Peptostreptococcus* BL, *Eubacterium limosum*, *Bacteroides praeacutus*, *Bacteroides* L, *Fusobacterium mortiferum* I, *F. naviforme*, *Clostridium innocuum*, *C. ramosum*, *Propionibacterium acnes*, *Ruminococcus flavefaciens*, *Ruminococcus* AT, *Peptococcus* AU-1, *Eubacterium* AG, -AK, -AL, -AL-1, -AN; *Bacteroides fragilis* ss. *ovatus*, -ss. *d*, -ss. *f*; *Bacteroides* L-1, L-5; *Fusobacterium nucleatum*, *F. mortiferum*, *Escherichia coli*, *Streptococcus morbillorum*, *Peptococcus magnus*, *Peptococcus* G, -AU-2; *Streptococcus intermedius*, *Ruminococcus lactaris*, *Ruminococcus* CO, *Gemmiger* X, *Coprococcus* BH, -CC; *Eubacterium tenue*, *Eubacterium ramulus*, *Eubacterium* AE, -AG-H, -AG-M, -AJ, -BW-1, *Bacteroides clostridiiformis* ss. *clostridiiformis*, *B. coagulans*, *B. oralis*, *B. ruminicola* ss. *brevis*, -ss. *ruminicola*, *Bacteroides splanchnicus*, *Desulfomonas pigra*, *Bacteroides* L-4, -W-1; *Fusobacterium* H, *Lactobacillus* G, and *Succinivibrio* A.


25. The composition of claims 23, or 24 wherein the predetermined microflora is in the form of a liquid culture of *Bacteroides* and *E. coli*.

26. The composition of any one of claims 23 to 25, in the form of an enema, enteric coated capsule or powder suitable for reconstitution.



# INTERNATIONAL SEARCH REPORT

International Application No. PCT/AU 89/00328

<b>I. CLASSIFICATION OF SUBJECT MATTER</b> (if several classification symbols apply, indicate all) 6				
According to International Patent Classification (IPC) or to both National Classification and IPC Int. Cl. <sup>4</sup> A61K 39/02, 39/08, 39/10, 39/11, 35/24				
<b>II. FIELDS SEARCHED</b>				
Minimum Documentation Searched 7				
Classification System	Classification Symbols			
IPC	A61K 39/02, 39/08, 39/10, 39/11, 35/24			
Documentation Searched other than Minimum Documentation to the Extent that such Documents are Included in the Fields Searched 8				
AU: IPC as above				
<b>III. DOCUMENTS CONSIDERED TO BE RELEVANT 9</b>				
Category*	Citation of Document, <sup>11</sup> with indication, where appropriate, of the relevant passages <sup>12</sup>	Relevant <sup>13</sup> Claim No		
X	AU,B, 53298/79 (526326) (NESTLE S.A.) 10 JULY 1980 (10.07.80) See page 2 lines 7-11, page 4 lines 20-27 and claims	(22-24,26)		
X	US,A, 3713836 (CARLSSON) 30 JANUARY 1973 (30.01.73) see column 1 lines 24-26, 40-44, column 3 lines 48-57	(22)		
X	US,A, 4335107 (SNOEYENBOS) 15 JUNE 1982 (15.06.82) See column 1 lines 43-52, column 2 lines 27,28, claims	(22,26)		
X	GB,A, 1271674 (NISSHIN FLOUR MILLING CO) 26 APRIL 1972 (26.04.72) See page 8 lines 1-4, 13-15, page 9 Example 1 and claims	(22,23,26)		
CONTINUED				
<p>* Special categories of cited documents: 10</p> <table style="width: 100%;"> <tr> <td style="width: 50%;"> <p>"A" document defining the general state of the art which is not considered to be of particular relevance</p> <p>"E" earlier document but published on or after the international filing date</p> <p>"L" document which may throw doubts on priority claim(s) or which is cited to establish the publication date of another citation or other special reason (as specified)</p> <p>"O" document referring to an oral disclosure, use, exhibition or other means</p> <p>"P" document published prior to the international filing date but later than the priority date claimed</p> </td> <td style="width: 50%;"> <p>"T" Later document published after the international filing date or priority date and not in conflict with the application but cited to understand the principle or theory underlying the invention</p> <p>"X" document of particular relevance; the claimed invention cannot be considered novel or cannot be considered to involve an inventive step</p> <p>"Y" document of particular relevance; the claimed invention cannot be considered to involve an inventive step when the document is combined with one or more other such documents, such combination being obvious to a person skilled in the art.</p> <p>"Z" document member of the same patent family</p> </td> </tr> </table>			<p>"A" document defining the general state of the art which is not considered to be of particular relevance</p> <p>"E" earlier document but published on or after the international filing date</p> <p>"L" document which may throw doubts on priority claim(s) or which is cited to establish the publication date of another citation or other special reason (as specified)</p> <p>"O" document referring to an oral disclosure, use, exhibition or other means</p> <p>"P" document published prior to the international filing date but later than the priority date claimed</p>	<p>"T" Later document published after the international filing date or priority date and not in conflict with the application but cited to understand the principle or theory underlying the invention</p> <p>"X" document of particular relevance; the claimed invention cannot be considered novel or cannot be considered to involve an inventive step</p> <p>"Y" document of particular relevance; the claimed invention cannot be considered to involve an inventive step when the document is combined with one or more other such documents, such combination being obvious to a person skilled in the art.</p> <p>"Z" document member of the same patent family</p>
<p>"A" document defining the general state of the art which is not considered to be of particular relevance</p> <p>"E" earlier document but published on or after the international filing date</p> <p>"L" document which may throw doubts on priority claim(s) or which is cited to establish the publication date of another citation or other special reason (as specified)</p> <p>"O" document referring to an oral disclosure, use, exhibition or other means</p> <p>"P" document published prior to the international filing date but later than the priority date claimed</p>	<p>"T" Later document published after the international filing date or priority date and not in conflict with the application but cited to understand the principle or theory underlying the invention</p> <p>"X" document of particular relevance; the claimed invention cannot be considered novel or cannot be considered to involve an inventive step</p> <p>"Y" document of particular relevance; the claimed invention cannot be considered to involve an inventive step when the document is combined with one or more other such documents, such combination being obvious to a person skilled in the art.</p> <p>"Z" document member of the same patent family</p>			
<b>IV. CERTIFICATION</b>				
Date of the Actual Completion of the International Search	Date of Mailing of this International Search Report			
15 NOVEMBER 1989	20 November 1989			
International Searching Authority	Signature of Authorized Officer			
Australian Patent Office	T NIZNIK 			

## FURTHER INFORMATION CONTINUED FROM THE SECOND SHEET

X	DE,A, 2134179 (SCHULER) 25 JANUARY 1973 (25.01.73) See claims	(22,23,26)
X	FR,M, 1275 (RENE ET AL) 2 MAY 1962 (02.05.62)	(22,23,26)
X	FR,M, 5528 (CARRAZ) 18 DECEMBER 1967 (18.12.67)	(22,26)
X	FR,M, 2427 (EURORGA S.A.) 1964 See page 1 lines 1-8, claims	(22,23,26)
X	FR,A, 2244464 (LABORATOIRES SEROZYM) 18 APRIL 1975 (18.04.75) See page 1 lines 1-3, lines 17-20 pages 3-4, examples, claims	
X	Patents Abstracts of Japan, C-46 page 141, JP,A, 54-35214 (NISSHIN SEIFUN K.K.) 15 MARCH 1979 (15.03.79)	(22,23,26)
	CONTINUED	

V. ☐ OBSERVATIONS WHERE CERTAIN CLAIMS WERE FOUND INSEARCHABLE 1

This international search report has not been established in respect of certain claims under Article 17(2)(a) for the following reasons:

1. ☒ Claim numbers 1-21, because they relate to subject matter not required to be searched by this Authority, namely:  
Methods of treatment of the human body by surgery or therapy.
2. ☐ Claim numbers ..., because they relate to parts of the international application that do not comply with the prescribed requirements to such an extent that no meaningful international search can be carried out, specifically:
3. ☐ Claim numbers ..., because they are dependent claims and are not drafted in accordance with the second and third sentences of PCT Rule 6.4 (a):

VI. ☐ OBSERVATIONS WHERE UNITY OF INVENTION IS LACKING 2

This International Searching Authority found multiple inventions in this international application as follows:

1. ☐ As all required additional search fees were timely paid by the applicant, this international search report covers all searchable claims of the international application.
2. ☐ As only some of the required additional search fees were timely paid by the applicant, this international search report covers only those claims of the international application for which fees were paid, specifically claims:
3. ☐ No required additional search fees were timely paid by the applicant. Consequently, this international search report is restricted to the invention first mentioned in the claims; it is covered by claim numbers:
4. ☐ As all searchable claims could be searched without effort justifying an additional fee, the International Searching Authority did not invite payment of any additional fee.

## Remark on Protest

- ☐ The additional search fees were accompanied by applicant's protest.  
☐ No protest accompanied the payment of additional search fees.

III. DOCUMENTS CONSIDERED TO BE RELEVANT (CONTINUED FROM THE SECOND SHEET)		
Category*	Citation of Document, with indication, where appropriate, of the relevant passages	Relevant to Claim No
A	FR,M, 2828 (NOUVEL) 2 NOVEMBER 1964 (02.11.64) see page 1 lines 1-34, claims	(22,23,26)
A	AU,B, 37580/85 (581198) (FARMOS O.Y.) 5 SEPTEMBER 1985 (05.09.85) See claim 1	(22)

ANNEX TO THE INTERNATIONAL SEARCH REPORT ON  
INTERNATIONAL APPLICATION NO. PCT/AU 89/00328

This Annex lists the known "A" publication level patent family members relating to the patent documents cited in the above-mentioned international search report. The Australian Patent Office is in no way liable for these particulars which are merely given for the purpose of information.

Patent Document Cited in Search Report		Patent Family Members			
<hr/>					
AU	526326				
<hr/>					
US	3713836	AT	296923	BE	747129
		CH	542281	DE	2010654
		FR	2068543	GB	1300446
		JP	50038682	IU	60508
		SE	371209	CA	933409
				ES	377347
				IT	1142151
				NL	167724
<hr/>					
US	4335107	AT	11641	DK	154927
		IE	42298	JP	1007049
				EP	6695
<hr/>					
GB	1271674	DE	1934652	FR	2014544
<hr/>					
FR	M 1275				
<hr/>					
FR	M 5528				
<hr/>					
FR	M 2427	IU	44002		

END OF ANNEX